### **ENDING IT ALL: EUTHANASIA OF REPTILES**

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### Introduction

"The greatness of a nation and its moral progress can be judged by the way its animals are treated" (Ghandi)

While this can obviously be applied to how animals are treated when they are alive it can also just as well be applied to how they are dealt with in death.

As every veterinarian and wildlife carer in the world knows it is impossible to save the life of every animal that is presented to us. Death, whether it is via natural means or euthanasia, is an inescapable fact.

The word "euthanasia" is derived from the Greek terms 'eu' meaning good and 'thanatos' meaning death and has thus been translated to mean "a gentle and easy death" or an "easy or painless death"... and that is the way it should be.

When the decision has been made to euthanase an animal it is our responsibility to ensure that it is done with the highest degree of respect, and with the emphasis on making the death as painless and distress free as possible.

#### Methods of Euthanasia

The euthanasia of reptiles and amphibians provides some unique challenges because of their anatomy and physiology. In order to overcome these challenges there are several factors to consider when selecting an appropriate method of euthanasia. These include:

- 1. The age of the animal e.g. foetus, very young, adult
- 2. The size of the animal.
- 3. Venous access. This may be difficult in some species or in animals suffering shock.
- 4. Animal handling and restraint. There may be safety concerns when dealing with venomous or very large reptiles. In addition there may be issues with exposing people, such as pregnant women, to anaesthetic gases.
- 5. Equipment available. Equipment may be required for restraint of some species in some situations (e.g. clear tubes for venomous snakes).
- 6. Tolerance and intolerance of certain species to particular agents. Amphibians and reptiles have a slow metabolism and have a high tolerance to hypoxia. This means that euthanasia methods that rely on anoxia may not be as effective in reptiles and amphibians when compared to other species such as mammals.
- 7. Experience of the person performing the euthanasia.
- 8. Access to a veterinarian

It is also important to appreciate that our understanding of amphibians' and reptiles' ability to feel pain and react to it is incomplete. While much research in this field is underway and our level of knowledge is ever increasing we must accept there is considerably more to learn.

All information provided in this presentation is based on the author's belief that euthanasia should be carried out by a veterinarian or at least by someone trained appropriately in the practice of euthanasia. If you are not trained and comfortable performing euthanasia then do not do it!

The euthanasia technique used should result in rapid loss of consciousness followed by cardiac or respiratory arrest with ultimately a loss of brain function. The methods used to euthanase reptiles and amphibians can be divided into 4 main areas and it is important to understand that many of them involve using a "two stage" process where the animal is sedated or anaesthetised prior to the euthanasia being performed. The methods are:

# Non-inhaled Agents

These methods of euthanasia involve chemical agents that are introduced into the body via means other than through the respiratory tract. It is vitally important to realise that many non-inhaled methods of euthanasia can result in a state of unconsciousness from which an animal can recover. As such it is important to use the "two stage" process as already mentioned and always confirm the animal is dead. These agents can be administered via:

i. Parenteral Injection.

Intravenous injection is one of the most rapid and reliable methods of performing euthanasia. It is by far the most desirable method if and when it can be performed safely and without causing fear or distress in the animal. Where this is not possible then the animal should be first sedated (e.g. intramuscular injection with drugs such as ketamine, tiletamine, medetomidine, propofol or alfaxalone) to allow easier intravenous access. Where intravenous administration is considered impractical or impossible (e.g. in very small animals) intracoelomic injection or injection into the subcutaneous lymph spaces and lymph sacs of a non-irritating chemical is acceptable.

# Agents that can be used include:

a. Barbiturates.

These act by depressing the central nervous system in descending order beginning with the cerebral cortex, with loss of consciousness progressing to anaesthesia, cessation of breathing and then cardiac arrest.

They act rapidly, induce euthanasia smoothly with minimal discomfort to the animal, are cheap and are easily accessible to veterinarians.

The most common barbiturate used for euthanasia is sodium pentobarbital. Trade names for this drug include Lethabarb® and Valabarb®. It is also often referred to as "the green dream". This is due to the fact that a green dye is added to it to distinguish it from all other injectable medications. In some countries a blue or red dye is added to it instead of green.

The standard dose of sodium pentobarbital for euthanasia is 60-100mg/kg.

When given via an intracoelomic injection it should be remembered that solutions such as these are very alkaline with a pH of around 11.0. Because of this they can be quite painful when given. For this reason it is best to dilute the normal concentrated solution (325mg/ml) to a solution that is 60mg/ml. This can be achieved by diluting 0.18ml of euthanasia solution with 0.82ml of saline or water for injection in a 1ml syringe. A small gauge needle (25-27 gauge) should also be used to minimise injected at a dose of 1ml per kg bodyweight. The table below gives a quick reference guide.

Body Weight (grams)	Amount to inject (ml)
	, , ,
≤10	0.01
20	0.02
50	0.05
70	0.07
100	0.1
200	0.2
300	0.3
400	0.4
500	0.5

There are reports of barbiturates being injected directly into the parietal eye in anaesthetised reptiles.

b. Potassium Chloride (KCl)

Although unacceptable when used in conscious animals, a rapid intravenous or intracardiac (IC) injection of KCl in an animal that is unconscious or under general anaesthesia is an acceptable way to induce cardiac arrest and death.

The dose of KCl is 75-150mg/kg rapidly IV or IC.

ii. Topical Application. This method tends to be slow and variable but has been used to euthanase animals with highly permeable skin such as amphibians. Agents used:

- a. Benzocaine hydrochloride.
  Application of 20% benzocaine hydrochloride gel in a 2cm x 2cm square approximately 1mm thick to the ventral abdomen of amphibians is an effective means of anaesthesia and euthanasia. It can result in a loss of reflexes in as little as 7 minutes and death in 5 hours.
- iii. Immersion. In some cases this is a suitable method of euthanasia for aquatic amphibians. Ideally the agent used will be non-irritating to the skin, eye, oral and respiratory tissue and will result in a rapid loss of consciousness.

## Agents used are:

a. Tricaine methanesulfonate (MS 222®)

While commercially available it is expensive if only using it on rare occasions, requires some special handling (as it requires mixing and buffering) and will deteriorate with time (it should be kept in the dark and refrigerated). It can be used for euthanasia of amphibians and some small reptiles however a secondary method should be used to ensure death.

It can be administered via a water bath, injected directly into the lymph sacs of amphibians or injected intracoelomically in amphibians and reptiles.

Prolonged immersion (as long as 1 hour) in a 5-10g/L water bath may be required to bring about death.

b. Isoeugenol (Aqui-S®)

Marketed as an anaesthetic agent for fish isoeugenol can be used to euthanase amphibians using an immersion bath. Amphibians can be immersed in the solution until no visible signs of life are seen but death should be confirmed or an adjunctive method used to ensure death.

#### c. Clove Oil

Cloves contain a number of essential oils including eugenol and isoeugenol. In human medicine clove oil has been used to treat tooth aches due to its local anaesthetic properties. Clove oil should be mixed with warm water first before adding it to the immersion bath. A concentration of 400mg clove oil per litre of water is required. Like isoeugenol amphibians can be immersed in the solution until no visible signs of life are seen but death should be confirmed or an adjunctive method used to ensure death

#### d. Alcohol

Immersion in alcohols such as ethanol can be used to euthanase small amphibians. They should be placed in a 10% solution until sedated and can then be placed in a 70% solution until dead.

iv. Oral administration. This method is regarded as impractical and of no use in reptiles and amphibians.

## Inhaled Agents

The suitability of any particular inhaled agent for euthanasia depends on the distress and/or pain experienced prior to the loss of consciousness.

Reptiles and amphibians have a great capacity to hold their breath and shunt their blood away from non-vital organs. This is particularly so for turtles and crocodiles. This can allow some species to survive for up to 27 hours when exposed to prolonged periods of hypoxia. As such the use of inhaled agents may result in prolonged times before a loss of consciousness occurs and death may not even occur.

Regardless of the species involved and the inhaled agent used a second, guaranteed lethal procedure should be performed to ensure death.

# Agents that can be used include:

- Inhaled anaesthetics.
   These include halothane, isoflurane and sevoflurane.
- b. Carbon dioxide.
   While this may be considered to euthanase amphibians and reptiles other methods are preferred. This is due to the fact that there is

potential lack of response to this method by many species, it requires prolonged exposure time and has the potential to cause severe distress and pain if not performed correctly. It is important that death is verified and assured by application of a secondary lethal procedure.

## Physical Methods

There are effectively two physical methods that can be used:

- i. Captive bolt or gunshot.
  - Crocodilians and other large reptiles can be euthanased by a penetrating captive bolt or gunshot delivered to the brain. There are diagrams of the heads of assorted species available showing recommended locations for such penetrative methods.
  - Obviously appropriate equipment and adequately trained personnel are required for this method.
- ii. Blunt force trauma to the head.
  - This method is acceptable when no other options are available but ideally alternate methods should be sought. It must be performed by a well-trained person and an additional method such as pithing promptly done to ensure death.

## Adjunctive Methods

As stated previously in many cases a "two stage" process of euthanasia is the preferred method of euthanasia in reptiles and amphibians. Many of the previously mentioned methods result in the loss of consciousness of an animal but not necessarily in its death. As such adjunctive methods are required to ensure the death of the animal. These include:

- i. Decapitation.
  - After an animal has been anaesthetised, decapitation using heavy shears is effective in small to medium sized animals.

It has been long assumed that stopping the blood supply to the brain causes a rapid loss of consciousness but this may not be the case in reptiles. Because the reptilian brain is tolerant of extremely low oxygen levels it is possible for the brain to function for a period of time after decapitation. Decapitation should be followed by pithing or another method of destroying brain tissue.

ii. Pithing.

Pithing is the process of inserting a sharp object in to the brain and using it destroy brain tissue. It should never be done on a conscious animal and should only be performed by trained individuals.

## **Some Comments About Freezing**

One of the most commonly suggested methods of euthanasing reptiles and amphibians is to simply place them in the freezer. Freezing, or more correctly, the use of hypothermia, is an inappropriate method of restraint or euthanasia for amphibians yet it is still widely practiced and is even listed as an appropriate method in some government-produced documentation.

The theory was that as reptiles are "cold blooded" simply cooling them down reduced their metabolism to a point where it just stopped. This is simply not true!

Hypothermic reptiles and amphibians are still able to function and feel pain but they are unable to demonstrate this pain either through behavioural (e.g. moving away) or physiological (e.g. increased heart rate) means.

But how would a freezing reptile be exposed to pain? Freezing results in the formation of ice crystals in tissues that cause extreme pain BEFORE the animal is dead.

It is also important to remember that there are certain species of amphibian that are able to survive freezing and in fact do so every year. Examples include Wood Frog (Lithobates sylvaticus) and the Western Chorus Frog (Pseudacris triseriata).

The only suitable methods of using freezing to euthanase an amphibian or reptile is if the animal is less than 7 grams in bodyweight and is placed directly in liquid nitrogen. Alternatively the animal must be anesthetised prior to being put into the freezer. Once an animal is thought to be dead it can be frozen as an extra measure to ensure death.

### **Confirmation of Death**

There are several ways of verifying death. These include:

1. Auscultation.

The animal should not be breathing and there should be no audible heartbeat.

2. Electrocardiograph (ECG).

There should be no detectable electrical activity from the heart.

3. Doppler.

This is a useful method for detecting a heartbeat or lack thereof.

4. Ultrasound.

Like Doppler an ultrasound machine is an effective way of detecting a heartbeat.

5. Pulse oximetry.

This involves determining the oxygen content of blood. Because of the anatomy of reptiles and amphibians it is of little practical use.

6. Visualisation

In certain animals such as smaller snakes, lizards and amphibians it may be possible to note the cessation of a heartbeat. Additionally the visualisation of respiration can be noted.

It is important to remember that amphibian and reptilian hearts can beat for up to several hours even after brain death. This is considerably longer than is seen with the mammalian heart.

# **Summary Table**

The information provided above is summarised in this table for use as a quick reference.

Agent/Method	Species Applied To	Dose	Comments
Non-Inhaled Agents	5		
Sodium pentobarbital	Reptile, Amphibian	60-100mg/kg IV (or IC in anaesthetised	Sedate the animal if required to

animals)	
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allow IV access

If given
intracoelomically
refer to previous
table for dilution
and dosage rates

		and dosage rates	
Potassium chloride	Reptile, Amphibian	75-150mg/kg rapidly IV or IC	Must only ever be given to an anaesthetised animal
Benzocaine hydrochloride	Amphibian	20% benzocaine hydrochloride gel in a 2cm x 2cm square approximately 1mm thick to the	Can take up to 5 hours for death to occur  Adjunctive method should
		ventral abdomen	be used in order to ensure death
Tricaine methanesulfonate (MS 222®)	Reptile, Amphibian	Intracoelomic injection in reptiles  Water bath,	Adjunctive method should be used in order to ensure death
(M3 222®)		injection into lymph sacs or intracoelomic for amphibians	
Isoeugenol (Aqui- S®)	Amphibian	Water bath	Adjunctive method should

			be used in order to ensure death
Clove oil	Amphibian	Water bath containing 400mg per 1L water	Adjunctive method should be used in order to ensure death
Alcohol	Amphibian	Place in 10% solution until sedated and then in 70% solution	Adjunctive method should be used in order to ensure death
Inhaled Agents			
Inhaled anaesthetics	Reptile, Amphibian		Reptiles, particularly turtles, breath hold and so very long exposure times may be required.
			Adjunctive method should be used in order to ensure death
Carbon dioxide	Reptile, Amphibian		Generally not recommended as a method of euthanasia for reptiles and

			amphibians
Physical Methods			
Captive bolt or gunshot	Reptile, Amphibian		Requires appropriate equipment and training
Blunt force trauma	Reptile, Amphibian		Adjunctive method should be used in order to ensure death
Adjunctive methods			
			Should only be performed on anaesthetised animal
Decapitation	Reptile, Amphibian		Should be immediately followed by pithing or other method to destroy the brain tissue
Pithing	Reptile, Amphibian		Should only be performed on anaesthetised animal
Freezing	Reptile, Amphibian	Animal less than 7 grams immersed in	Is not a suitable method of euthanasia

#### References

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- 4. Cooper J. Humane Euthanasia and Post-Mortem Examination in BSAVA Manual of Reptiles 2nd Edition. Girling S and Raiti P: pp 168-183. British Small Animal Veterinary Association.

**Dr Shane Simpson :** As a veterinarian with a special interest in reptiles and amphibians, Dr Shane Simpson is probably better known as "The Reptile Doctor". Each year he treats over 2000 pet and wild reptiles at Karingal Veterinary Hospital located in the south-eastern suburbs of Melbourne.

Dr Simpson regularly lectures to fellow veterinarians, veterinary nurses and reptile keepers on assorted topics relating to reptile and amphibian medicine and surgery. In addition he regularly contributes articles and other material to reptile magazines, veterinary journals and websites as well as being an active participant on several online forums and Facebook pages dedicated to reptiles and amphibians. He is a consultant veterinarian for a number of reptile shops and aquariums, wildlife shelters, reptile demonstrator businesses, wildlife parks and large commercial collections. Dr Simpson is a member of the Australian Veterinary Association, The Unusual and Exotic Pet and Avian Vet Association, the Association of Reptile and Amphibian Veterinarians and the Victorian Herpetological Society.